

CYTOTOXICITY EVALUATIONS OF CARBON DOTS WITH DIFFERENT SURFACE CHARGE

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Abstract

Carbon dots (CDs) are a new class of the fluorescent nanoprobe which have been found promising for bio-applications. Surface functionalization plays a critical role in the cytotoxicity determination of carbon dots and is a very important issue. One of the main factors which affect the cytotoxicity is a surface charge as influences cellular/intracellular tracking of nanomaterials. Thus, a comparative study of CDs with different surface charges was performed on standard cells mouse fibroblasts (NIH-3T3). We tested pristine carbon dots derived from candle soot with the same CDs functionalized with polymers. Polyethylenimine (PEI) and polyethylenglycol (PEG) have been used to obtain different surface charges of CDs (PEI - positive charge, PEG neutral charge); pristine CDs bear negative charge by themselves. Changes of viability were measured by MTT assay, oxidative stress was determined through ROS analysis, and the cell cycle profile was obtained by flow cytometry analysis. Morphology changes in the cells caused by incorporated carbon dots were observed by light microscopy. From cytotoxicity measurement, we identified that the surface chemistry affects viability and oxidative stress. The most toxic response was induced by positively charged PEI-carbon dots at a concentration of 50 $\mu\text{g. mL}^{-1}$. Viability of the cells loaded by PEGylated carbon dots did not decrease significantly up to 300 $\mu\text{g. mL}^{-1}$. The bare CDs stimulated proliferation at the low concentration until 100 $\mu\text{g. mL}^{-1}$ and then viability went down with increasing concentration (IC₅₀ value was found at 300 $\mu\text{g. mL}^{-1}$). ROS kinetic study has shown that the highest oxidative stress occurred in cells labeled with pristine carbon dots. PEGylated CDs did not cause a considerable ROS level compared to the control cells. From the cell cycle analysis, we evaluated the arrests in the individual phases. PEGylated carbon dots did not display any abnormality in proportion of the cell cycle. Cells treated with 50 $\mu\text{g. mL}^{-1}$ of the PEI CDs tend to be arrested in G₀/G₁ and the G₂/M phase became relatively more populated than the S phase. Pristine carbon dots did not affect the cell cycle significantly at a concentration of 50 $\mu\text{g. mL}^{-1}$ but at 100 $\mu\text{g. mL}^{-1}$, the G₂/M arrest occurred. The arrest in G₀/G₁ occurred at the 350 $\mu\text{g. mL}^{-1}$ treatment. We carried out the comprehensive cytotoxicity study and found out a different biocompatibility of carbon dots depending on the surface chemistry.

Keywords: Carbon dots, surface chemistry, cytotoxicity

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